2908-Pos Board B13
The Short-Time Dynamics of Proteins Near Native State Conditions Signal to Robust Mechanisms Accessible at Long Times
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A number of recent computational studies have indicated that a robust pattern of motions is encoded by the particular architecture of a protein. These patterns have been shown to be of relevance to functional motions that occur on the much slower time scale of microseconds or longer. In addition, experimental data derived from NMR relaxation and other motion-containing NMR parameters as well as computational data from MD simulations indicate that the relative movements of residues at short times exhibit discernable correlations with those observed at long times. This intriguing issue was explored in the present work by conducting systematic molecular dynamics simulations of various durations, varying in the range 1 ns $\leq t_{tot} \leq$ 0.4 μs. While the observed amplitudes of motions are generally confined to 1 Å in short simulations with a general increase to 2-3 Å in the long ones, strikingly, the residue displacement distributions (away from equilibrium positions) exhibit comparable patterns. Careful examination of the relationship between the size of observed motion and the duration of simulations revealed a stretched exponential dependence of the the form y μ $x^{0.26}$, with x as representing the ratio of simulation lengths and y the ratio of the motional amplitudes, consistent with the observations made by Scheraga and coworkers.[1]
1. Senet P, Maisuradze GG, Foulie C, Delarue P, Scheraga HA. How main-chains of proteins explore the free-energy landscape in native states. *Proc. Natl. Acad. Sci. U. S. A* (2008) 105: 19708-19713.

2909-Pos Board B14
Allosterism in Muts Proteins: How DNA Mismatch Recognition Signals Repair
Susan N. Pieniazek, Manju M. Hingorani, David L. Beveridge.
Allosteric communication in multi-domain protein architectures is crucial in complex biological processes such as DNA mismatch repair (MMR). MutS proteins initiate MMR through recognition of mismatch DNA and signaling downstream repair. Mismatch recognition by MutS is followed by a marked decrease in the rate of ATP hydrolysis and DNA binding affinity. However, the nature of the coupling between the ATPase sites and DNA-binding site ~70 Å away remains a mystery, mainly because MutS is a relatively large dimeric protein.
Vast networks of interactions serve as support for protein structure. In MutS proteins, organized networks couple DNA mismatch affinity and ATP hydrolysis. We have performed all-atom molecular dynamics simulations (150 ns) on ATP-bound and ATP-free MutS complexes with mismatch DNA to understand the effect of ATP binding on structural networks connecting the ATPase sites and DNA-binding site. In particular, the eigenvectors of the correlation matrix reveal networks of mutually correlated residues, which are related to the paths of allosteric communication in MutS. Overall, several specific MutS structural components thought to be involved in allosteric coupling between DNA-binding and ATPase domains are present in the four largest eigenvalues: A) the involvement of domain II, which resides at the connection between domains V and III, appears in most of the correlations, especially in eigenvector 1; B) From eigenvector 1, MutS rigid domains III and V move together as a unit supporting their key role in the transmission of the signal from the, more flexible, DNA binding region to the ATPase site; C) the salient networks of correlated residues from eigenvector 4 (MutS-ATP-DNA) and eigenvector 3 (MutS-DNA) predominantly involve highly conserved long α-helical levers (IIIb and IV).

2910-Pos Board B15
Dynamics Encode Dynamically Committed and Uncommitted States in Protein Kinase A
Larry R. Masterson, Gianluigi Veglia.
The catalytic subunit of protein kinase A (PKA-C) is a promiscuous enzyme which phosphorylates a number of proteins in the cell. Our current view of the atomic details of substrate recognition has been derived from crystal structures with standard, largely non-physiologically relevant substrates. Herein, we present the full amide backbone dynamics of PKA-C bound to a physiologically relevant substrate (phospholamban peptide, PLN_{1-20}) and an inhibitor (protein kinase inhibitor, PKI_{5-24}). We found a direct correlation of decreasing dynamics in conserved residues of PKA-C as catalytic efficiency decreased. Strikingly, this decreased dynamics was also accompanied by increased thermal stability and unfavorable binding entropy. The dynamic character of the enzyme in the presence of a substrate or inhibitor is highlighted by residues that line the active site at the conserved glycine-rich, activation, and peptide positioning loops. These residues seem to display similar dynamics as the enzyme recognizes phosphorylatable substrates. The dynamics are severely attenuated when the enzyme binds PKI_{5-24}, particularly under high Mg^{2+} when inhibition is increased 100-fold. The necessity for concerted motions between the loops that line the active site opens the possibility for an evolutionary conserved role of dynamics which are encoded at spatially distinct regions of PKA-C. Molecular dynamics simulations support the observation that the conformational sampling of the enzyme is restricted by inhibition and that its energy landscape becomes well defined. We propose that ligand binding can induce either dynamically committed states in the enzyme that are driven by favorable conformational entropy, or dynamically uncommitted states that are driven by favorable enthalpy. The influence of dynamics on substrate recognition and the slow-step of catalysis will also be discussed.

2911-Pos Board B16
Molecular and Structural Insight for the Role of Key Residues of Thrombospondin-1 and Calreticulin in Thrombospondin-1- Calreticulin Binding
Qi Yan, Joanne E. Murphy-Ullrich, **Yuhua Song**.
Thrombospondin-1 (TSP1) binding to calreticulin (CRT) on cell surface signals focal adhesion disassembly. Residues of Lys 24 and 32 in TSP1 and amino acids 24-26 and 32-34 in CRT are critical for TSP1-CRT binding (Faseb J, 22:3968, 2008; J Biol Chem, 268:26784, 1993). This study investigated molecular and structural basis for the effect of these key residues in TSP1 and CRT on TSP1-CRT binding. Based on a validated TSP1-CRT complex structure (Biochemistry, 47:3685, 2010), we adopted steered molecular dynamics simulations to determine the effect of the mutations of these key residues on TSP1-CRT binding and validated the simulation results with experimental observation. We further performed 30 ns molecular dynamics simulations for wild type TSP1, CRT, TSP1 K24A&K32A mutant and mutant CRT (residues 24-26 & 32-34 mutated to Ala) and studied the conformational and structural changes of TSP1 and CRT by the mutations of the critical residues. Results showed that mutations of residues 24 and 32 to Ala in TSP1 and of amino acids 24-26 and 32-34 in CRT to Ala result in a shortened β strand in the binding site, decreased hydrogen bond occupancy for β strand pairs that are located within or near the binding site, increased conformational flexibility of the binding site, a changed degree of dynamic correlated motion between the residues in the binding site and the other residues in protein, and a changed degree of overall correlated motions between the residues in protein. These changes could directly contribute to the loss or reduced binding of TSP1-CRT complex and further affect TSP1-CRT binding induced cellular activities. Results from this study provide molecular and structural insight for the role of the critical residues in TSP1 and CRT in TSP1-CRT binding.

2912-Pos Board B17
Structural and Dynamic Effects Due to Glycation on Cholinesterases
César Millán-Pacheco, Benjamín Pérez-Aguilar, Noé Salinas-Arreortua, Eduardo Jardón-Valadez, José Luís Gómez-Olivares.
Diabetes mellitus is one of the most common diseases in the world. Currently around 3% of the global population suffers from diabetes and this is expected to double by 2030. During hyperglycemic episodes, protein glycation may occur on lysine residues. Decrease of biological activity as a direct effect of glycation has been shown on calmodulin, superoxide dismutase and calcium ATPase on human erythrocytes. In this work, we evaluated the structural and dynamical effects due to lysine glycation on human acetylcholinesterase and butyrylcholinesterase. Four probable sites were tested and analyzed for each cholinesterase by molecular dynamics simulations, using the CHARMM27 forcefield in NAMD at 298K, for 25ns on each model. The cumulative effect of each site was studied comparing it to the native, unglycated protein structure. The structural and dynamic consequences due to lysine glycation on the conformation of the choline binding site are reported.
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2913-Pos Board B18
Dynamics of Conformational Heterogeneity Within the Michaelis Complex of Lactate Dehydrogenase
Ruel Z. Desamero, Beining Nie, Nick Zhadin, Hua Deng, Robert Callender.
An enzymatic reaction involves the diffusion-controlled formation of an encounter complex between the protein and its substrate followed by the appropriate structural and dynamical arrangements producing Michaelis complex capable of product formation. In forming the Michaelis complex, the binding pocket is substantially rearranged: protein flaps or loops often close over the bound ligand, the binding pocket is desolvated, and catalytically important residues are brought into contact with the substrate. We probed the transient events associated with the binding of oxamate, a substrate mimic, to lactate dehydrogenase isolated from Bacillus stearothermophilus (bsLDH) using temperature jump (T-jump) relaxation techniques. T-jump relaxation monitors the